

Studies on Ambrosia, I. The Inheritance of Floral Types
in the Ragweed, *Ambrosia elatior* L.

K. L. JONES



A dissertation submitted in partial fulfillment
of the requirements for the degree of Doctor
of Philosophy in the University of Michigan.

Reprinted from

"THE AMERICAN MIDLAND NATURALIST"

Vol. 17, No. 4, pp. 673-699, July, 1936

The University Press
Notre Dame, Ind.



The American Midland Naturalist

Published Bi-Monthly by The University of Notre Dame, Notre Dame, Indiana

VOL. 17

JULY, 1936

No. 4

Studies on *Ambrosia*, I.

The inheritance of floral types in the ragweed, *Ambrosia elatior* L.¹

K. L. Jones

Introduction

There is disagreement over the status of the species *Ambrosia elatior* L. and *A. artemisiifolia* L. This is partly because of their treatment in the more commonly used manuals: Gray's Manual, for example, mentions only the latter, whereas, in Britton and Brown, *Ambrosia elatior* L. is described and *A. artemisiifolia* L. is given as a synonym. The writer has minutely examined some 30,000 individuals of the lesser ragweed, grown in his cultures the past six years at the University of Michigan Botanical Gardens. The characteristics which have been used to establish these species (Linnaeus, 1753; Rydberg, 1922) are largely quantitative, and in culture, are too variable to justify any segregation. Rydberg (1922) for example, restricts *A. artemisiifolia* L. to the Atlantic seaboard and makes *A. elatior* L. the ubiquitous species. The contrasting characteristics stated by this author are:

Ambrosia elatior L.: Stem 3-10 dm. high; leaves bipinnatifid or the upper less divided; staminate involucre 3 mm. wide; body of fruit 3 mm. long.

Ambrosia artemisiifolia L.: Stem 5-10 dm. high; leaves pinnatifid or the upper simple; staminate involucre fully 4 mm. wide; body of fruit 2.5-3 mm. long.

The plants which the present author grew from seed obtained locally at Ann Arbor, Michigan, and which would be *A. elatior* L. according to Rydberg, were characterized as follows: Stem 0.5-19.5 dm high; leaves usually bipinnatifid but ranging from entire to tripinnatifid; staminate involucre 2-20 mm wide; body of fruit 1.5-5 mm. long. The offspring of one plant may exceed the total range of variability in all those characteristics which have been used to discriminate between the two species. Consequently, the writer believes that the name *Ambrosia elatior* L. had better be retained since it has priority of position in the Species Plantarum.

The occurrence of aberrant floral types in *A. elatior* L. has long been known to field botanists, and this information has filtered into most manuals. Three floral types occur: (1) Normal type (Figs. 1 and 2); monoecious; pistillate heads borne laterally in leaf axils, staminate heads in terminal racemes. (2) Intermediate type (Figs. 3 and 4); monoecious; pistillate heads

¹ Papers from the Department of Botany, University of Michigan, number 510.

borne laterally and also in racemes where they accompany staminate or androgynous heads. (3) Pistillate type (Figs. 5 and 6); plant entirely pistillate: pistillate heads borne laterally and in racemes. There are no staminate individuals. All plants bear pistillate heads laterally and hence the types differ only in their terminal inflorescences.

The following description of the floral types is based upon a study of *A. elatior* L. growing wild in the vicinity of Ann Arbor, Michigan, where there is a remarkable array of interesting forms; from my cultures, which were mostly of local origin, but also contained plants from Florida, North Carolina and Arkansas; from herbarium specimens in the University of Michigan Herbarium, and the Swan-Myers Herbarium, the latter furnished through the kindness of O. C. Durham.

I. NORMAL TYPE. (Monoecious form 1 and 2).

Locally, at Ann Arbor, over 95% of *A. elatior* L. plants are of the normal type. (Figs. 1 and 2). From the information I have been able to obtain, this type is always predominant throughout the range of the species. In the normal type the distribution of the flowers is roughly like that of *Zea Mays*. The staminate flowers are borne in terminal inflorescences and the pistillate flowers are produced laterally. The minute flowers are in heads; the terminal inflorescence is really a raceme of heads. A staminate head consists of 20-30 flowers, partially concealed within a nodding, saucer-shaped involucre of united bracts which is about 3 mm. wide, and is attached to the rachis by a stalk less than 3 mm. long. At times the staminate racemes are compound. A staminate flower (Fig. 7) has a perianth of one whorl, which is funnelform, and five toothed; the five anthers are distinct and surround a vestigial pistil which is uniformly cylindrical except for a dilated tip.

Pistillate heads (Fig. 8) are in small groups, sessile and erect in the axils of leaves, bracts or at the base of staminate racemes. The involucre of united bracts fits tightly against the single pistil which it harbors, except for the long, forked style which extends through its subulate beak. There are never stamens nor rudiments of any parts of a staminate flower within a normal pistillate head of this type. The body of the involucre fruit is obovoid, about 3 mm. long, with a series of 4-10 short spines. The pericarp is black and rigid, and contains one relatively large seed in which the food is stored in cotyledons.

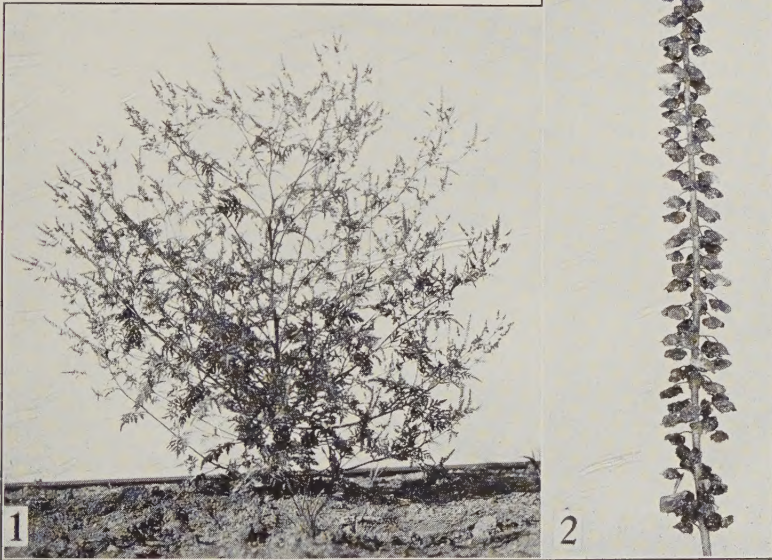
II. INTERMEDIATE TYPE. (Monoecious form 3).

The intermediate type (Figs. 3 and 4) differs from the normal type in that the pistils, in addition to being produced laterally, occur also in the terminal racemes along with staminate flowers. In the distribution of pistils in the terminal inflorescences, this type has been observed to range from individuals which would be pistillate but for one staminate flower in one androgynous head, to individuals which would be normal but for one pistil in one androgynous head. It is even possible that intermediates border still nearer the normal and pistillate types, because, in prepared slides a pistil has been

found to harbor a functional anther in its style (Fig. 9) and a staminate flower was sectioned in which a free ovule was present in place of a stamen (Fig. 10.) These examples are similar to the interesting findings of Yampolsky (1934) on *Mercurialis*. A list of intermediate types could be given which would almost satisfy all the possible numerical combinations and positions of stamens and pistils, in individuals bearing scores of racemes each of which

Fig. 1. Normal type; monoecious form 1 or 2.

Fig. 2. Normal type; monoecious form 1 or 2; raceme of staminate heads.



might contain 150 heads with an average of 20 flowers per head. The following list of types, therefore, while tedious, is but a skeleton classification.

1. Plants entirely pistillate but for one androgynous head.

Such androgynous heads, which yield functional pollen and seed, are not confined to plants that are predominantly pistillate. The involucre may be staminate, pistillate, or

a bizarre combination of these two. The head consists of staminate flowers and generally free, i.e. non-involucrate pistils. If the involucre is pistillate or nearly so, the ovary will be at least partially inclosed. The actual and relative number of staminate flowers and pistils within a head is variable. The following record of dissections describes a few representative types of androgynous heads:

- a. Involucre staminate; seven normal staminate flowers, one pistil.
- b. Involucre staminate; seven staminate flowers, two of which were contabescent and growing out of an ovary which set seed.
- c. Involucre staminate; one staminate flower, one pistil. Staminate flower produced functional pollen and grew from an aborted ovary.
- d. Involucre sectorially staminate and pistillate; one pistil with style through beak but the other half of the involucre saucer-shaped holding two staminate flowers.
- e. Involucre pistillate; two pistils with their styles extending through beak. Nine staminate flowers entirely concealed within involucre.

A confusing factor in the study of the intermediate type is the occasional lack of correlation between the form of the involucre and the floral structures contained therein. (see par. "e" above.) An involucre of staminate type may contain one to three functional pistils and no staminate structures. An involucre of the pistillate type, fully developed with spines and beak, may completely incase a dozen staminate flowers and bear no pistil; here, the pollen may never escape even though the anthers dehiscence normally.

2. Plants entirely pistillate but for one staminate head. This staminate head is usually terminal, although perhaps only because in such a position it is least likely to be overlooked.

3. Plants predominantly pistillate; a few staminate heads.

4. Plants having approximately the same number of staminate and pistillate heads and the two irregularly distributed.

5. Plants having approximately the same number of staminate and pistillate heads and the two occurring in segments of the racemes in serial alternation.

6. Plants with predominantly staminate racemes; a few pistillate heads present.

7. Plants with staminate racemes but for one pistillate or androgynous head.

III. PISTILLATE TYPE. (Figs. 5 and 6).

Plants of this type bear only pistillate heads laterally in the leaf axils and terminally in long racemes. In adverse environments one may find plants which are pistillate because they are so stunted that no racemes develop. These individuals may be potentially normal, intermediate or pistillate.

Methods

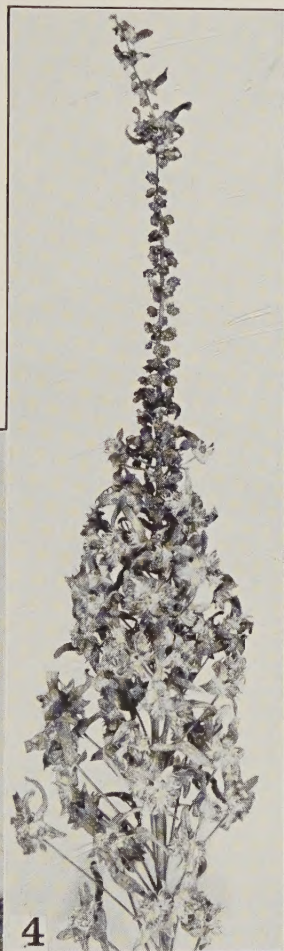
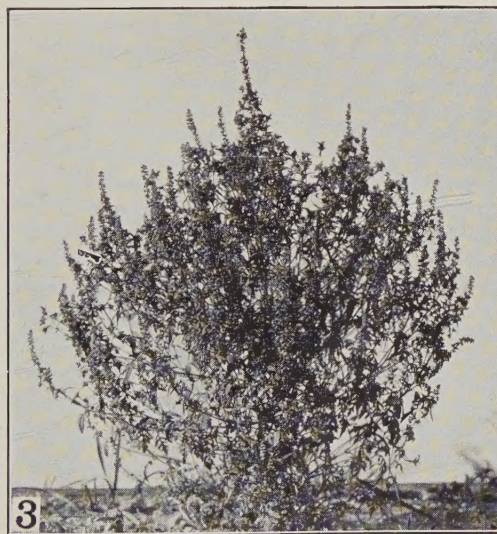
The writer began his studies of the sex condition in *Ambrosia elatior* L. during the summer of 1929. Previous to this the only work on the problem was an experiment by Garner and Allard (1920) in their well known investigations on photoperiodicity. Plants growing in the environs of Ann Arbor were examined and these data contributed to the classification of the floral types just described. The distribution in nature suggested that the genetic factors were "differential" (Sharp, 1924) and the environmental factors "conditional" in the development of the unlike types. Often normal, intermediate

and pistillate plants were in such close proximity that their roots were intermingling. Wherever the species occurred in any profusion the three sex types were present and the normal type predominated.

The initial collection of seed was necessarily from wild plants. The achenes are too numerous to be collected in the field; Stevens (1932) reported 3,380 on a representative plant. Collecting was done by removing entire

Fig. 3. Intermediate type; monoecious form 3.

Fig. 4. Intermediate type; monoecious form 3;
raceme of pistillate and staminate heads.



plants and storing them individually in large paper bags. After a specimen had dried it was crushed in the bag and winnowed over an electric fan to secure the seed free from leaf fragments.

Neither freshly harvested nor old seed will germinate without special treatment. Davis (1930) reported that *Ambrosia trifida* seed germinate if

after-ripened at 5 degrees C. one to two months. This method was satisfactory for *A. elatior*. The seed must be kept moist while after-ripening or there will be no germination. Sowings were made in sterilized soil. Germination was highest at 35 degrees C. Each seedling was transferred to a separate pot and labelled. If plants were later grown in the field the label accompanied the plant. The importance of thus avoiding contamination in growing a common weed is obvious.

Controlled pollination is impossible in the field because the plants are wind-pollinated, ubiquitous and the flowers are minute and inconveniently distributed. Seed will not set if the entire plant is bagged with cellophane. Consequently plants must be grown out of season in the greenhouse. Unfortunately *A. elatior* does not thrive under these conditions. The plants require strong light, a long photoperiod and a high temperature. They are very susceptible to white fly and thrips. If grown in the winter most individuals will produce few or no racemes and one is fortunate to secure a dozen viable seed from a plant.

Sowings in early March gave the best results. For example, cultures planted March 10th flowered from April 30th to July 31st. Pollination of ragweeds in nature, in Ann Arbor, begins the third week in August. In making crosses the plants used as female parents were kept emasculated, which requires daily examination, and placed in contingent rows with pollen parents of a given type. Contamination was obviated by maintaining but one type as a pollen parent a season or by utilizing the greenhouses at the Botanical Gardens and at the University of Michigan campus, the two being over a mile apart.

Results

a. THE GENETICAL BEHAVIOR OF PISTILLATE PLANTS.

F_1 open-pollinated.

The progeny of wild, open-pollinated pistillate plants were cultured in the field. All the seed produced by a plant were sown; each culture was derived from a single plant. The results are given in Table 1.

TABLE 1
 F_1 progeny of open-pollinated, pistillate plants.

Culture	No. Plants	No. N	No. I	No. P	%N	%I	%P
23P	531	29	115	387	5.4	21.6	72.8
96P	97	21	51	25	21.6	52.5	25.7
40P	806	239	360	207	29.6	44.6	25.6
33P	325	92	123	110	28.3	37.8	33.8
150P	642	218	184	240	33.9	28.6	37.3
Total	2401	599	833	969	24.9	34.6	40.3

N; normal type, monoecious form 1 or 2.

I; intermediate type, monoecious form 3.

P; pistillate type.

Each of the cultures was derived from one seed parent; e.g. 23P was produced by pistillate plant number 23.

An examination of Table 1 shows that in every instance the offspring of pistillate plants consisted of normal, intermediate and pistillate individuals. The frequency of each type was extremely variable but the normal type was never predominant.

In addition to the foregoing, seventeen cultures (2,690 plants) of similar origin were grown from which the writer secured incomplete data because he was not yet inured to certain inconveniences concomitant with a study of ragweed by one sensitized to its pollen. All of these cultures contained the three sex types; normal plants comprised from 6-30% of each culture.



Fig. 5. Pistillate type.

Fig. 6. Pistillate type; raceme of pistillate heads.

F_2 open-pollinated.

Pistillate plants were selected from the F_1 cultures 23P and 81P because they had given the highest percentage of the pistillate type. Culture 81P was among those from which data were incomplete; it consisted of over 50% pistillate plants. The F_2 cultures derived from these are presented in Table 2.

TABLE 2

F₂ progeny of open-pollinated, pistillate plants.

Culture	%Germ	No. plants	No. N	No. I	No. P	%N	%I	%P
23P-1P	84.4	201	21	112	68	10.4	55.7	33.8
-2P	43.2	104	47	53	4	45.1	50.9	3.8
-4P	50.8	112	32	54	26	28.5	48.2	23.2
-5P	59.2	137	11	43	83	8.0	31.3	60.5
-6P	76.8	142	22	80	40	15.4	56.3	28.1
-8P	69.2	82	3	32	47	3.6	39.0	57.3
-10P	47.2	104	32	40	32	30.7	38.4	30.7
-11P	75.2	165	14	97	54	8.4	58.7	32.7
-12P	62.8	148	22	74	52	14.8	50.0	35.1
-13P	48.4	109	4	46	59	3.6	42.2	54.1
-14P	55.2	139	7	68	64	5.0	48.9	46.0
-23P	19.6	49	2	26	21	4.0	53.0	42.8
-26P	80.4	170	3	69	98	1.7	40.5	57.6
-28P	71.2	107	7	26	74	6.5	24.2	69.1
-32P	87.2	183	36	58	89	19.6	31.6	48.6
-33P	70.8	166	22	72	72	13.2	43.3	43.3
-35P	82.4	198	115	79	4	58.0	39.8	2.0
-36P	84.4	193	60	83	50	31.0	43.0	25.9
-37P	84.4	199	13	46	140	6.5	23.1	70.3
-38P	68.4	166	45	63	58	27.1	37.9	34.9
-39P	72.8	172	7	47	118	4.0	27.3	68.6
Total	66.3	3046	525	1268	1253	17.2	41.6	41.1
81P-4P	58.4	122	12	49	61	9.8	40.1	50.0
-6P	91.6	219	26	95	97	11.9	43.5	44.4
-8P	63.2	154	24	62	68	15.5	40.2	44.1
Total	71.0	494	62	206	226	12.5	41.7	45.7
All	67.0	3540	587	1474	1479	16.5	41.6	41.7

The F₂ results were comparable in many respects to the F₁. Three sex types appeared in each F₂ culture. The normal type was predominant in only one culture, 23P-35P. The frequency of each sex type was extremely variable with the following ranges: normal, 1.7%-58.0%, intermediate, 23.1%-58.7%; pistillate, 2.0%-70.3%. None of the cultures had as high a percentage of pistillate plants as the F₁ parent culture; however six cultures had a lower percentage of the normal type. The percentage of intermediate plants was higher in every F₂ culture.

F₃ open-pollinated.

Pistillate plants were selected from the F₂ culture, 23P-26P, which had given the lowest percentage of the normal type and from 23P-38P in which the sex types were present in approximately equal proportions. The F₃ cultures derived from these are given in Table 3.

TABLE 3
F₃ progeny of open-pollinated, pistillate plants.

Culture	No. Plants	No. N	No. I	No. P	% N	% I	% P
23P-26P-1P	99	37	34	28	37.3	34.3	28.2
-2P	99	27	43	29	27.2	43.4	29.2
-3P	98	34	39	25	34.6	39.7	25.5
-4P	95	24	36	35	25.6	37.8	36.8
-5P	97	9	38	50	9.3	39.1	51.5
Total	488	131	190	167	26.8	38.9	34.2
23P-38P-1P	91	13	42	36	14.2	46.1	39.5
-2P	96	35	43	18	36.4	44.7	18.7
-3P	94	37	39	18	39.3	41.4	19.1
-4P	39	11	17	11	28.2	43.5	28.2
5P	49	13	14	22	26.5	28.5	44.8
Total	369	109	155	105	29.5	42.0	28.4
All	857	240	345	272	28.0	40.2	31.6

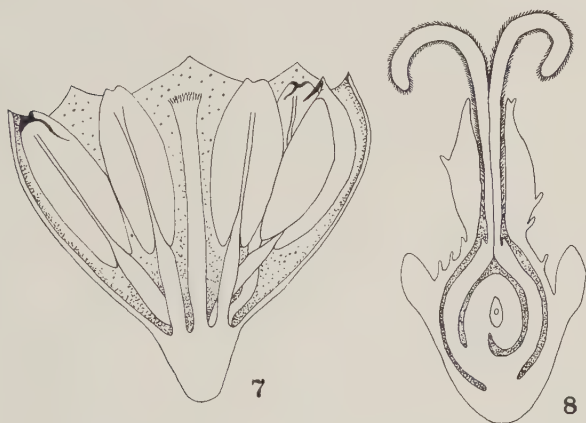


Fig. 7. Staminate flower; five stamens and a vestigial pistil.

Fig. 8. Pistillate head in longitudinal section.

Again, as in the F₁ and F₂, all three sex types were present in each culture. However, the aggregate results show a decided decrease in pistillate plants and an increase in the normal type. A comparison of individual cultures indicates extreme variation. Derivatives of 23P-26P always yielded a higher percentage of normal and a lower percentage of pistillate plants than the F₂ parent, whereas, cultures derived from 23P-38P varied in both directions for each sex type compared with the F₂. Selection for an increase in the frequency of pistillate plants under open-pollination was ineffectual; the percentage of pistillate plants decreased in succeeding generations and the percentage of normal plants increased.

The pistillate type in controlled crosses.

Pistillate plants of the F_2 generation of strain 23P were crossed with a true-breeding normal strain, 9N, and also with intermediate segregates of 23P. These two crosses were repeated with pistillate plants of the F_3 generation of strain 23P. The results are given in Table 4.

TABLE 4
Pistillate plants crossed with a true-breeding normal strain, 9N, and with intermediate plants.

Culture	No. Plants	No. N	No. I	No. P	%N	%I	%P
P $\times$ 9N	235	42	95	98	17.8	40.4	41.7
Repeated	40	8	14	18	20.0	35.0	45.0
Total	275	50	109	116	18.1	39.6	42.1
P $\times$ I	274	44	114	116	16.0	41.6	42.3
Repeated	96	12	42	42	12.5	43.7	43.7
Total	370	56	156	158	15.1	42.1	42.7

These data indicate that the throwing of three sex types by a pistillate plant is not conditioned by the sex type of the male parent. It is peculiar that the total results from these two crosses should be so similar when the frequency of sex types under open pollination has been extremely variable. Pistillate plants have also been crossed with the unstable normal segregates of 23P, monoecious form 2, but the offspring have not as yet been grown.

2. THE GENETICAL BEHAVIOR OF THE NORMAL TYPE.

F_1 open-pollinated. (Table 5).

TABLE 5
 F_1 progeny of open-pollinated, normal plants; monoecious form 1.

Culture	No. Plants	No. N	No. I	No. P	%N	%I	%P
*9N	186	186	0	0	100	0	0
100N	90	90	0	0	100	0	0
*101N	1110	1108	2	0	99.8	0.1	0
102N	99	99	0	0	100	0	0
103N	99	99	0	0	100	0	0
104N	99	99	0	0	100	0	0
*105N	25	25	0	0	100	0	0
*106N	61	61	0	0	100	0	0
107N	104	104	0	0	100	0	0
108N	97	96	1	0	98.9	1.0	0
109N	100	99	1	0	99.0	1.0	0
*110N	58	58	0	0	100	0	0
*112N	16	16	0	0	100	0	0
113N	100	100	0	0	100	0	0
114N	99	99	0	0	100	0	0
02N	97	97	0	0	100	0	0
04N	86	86	0	0	100	0	0
Total	2526	2522	4	0	99.84	0.15	0

*All seeds produced by the parent plant were sown.
Each culture was derived from the seed of one plant.

All cultures, except O2N and O4N, were derived from seed collected in the vicinity of Ann Arbor. Culture O2N was grown from seed collected at Fayetteville, Arkansas, through the kindness of Professor Delbert Swartz. The original seed in this case were not from one plant; from them 232 plants were grown, 98.7% normal and 1.28% of intermediate type. Culture O2N represents the offspring of one of these normal plants. Cultures O4N came from Tallahassee, Florida, from seed collected through the kindness of Professor R. M. Harper. A portion of the original seed yielded 160 plants all of the normal type. Culture O4N represents the offspring of one of these normal plants.

The data in Table 5 show that normal plants, which are so called because they are the predominant type in nature, breed quite true. The very few in-

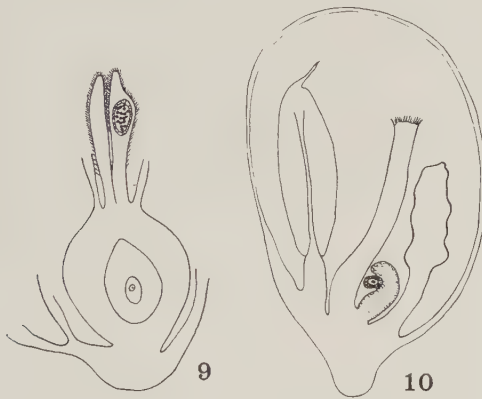


Fig. 9. Pistil with anther in style.

Fig. 10. Staminate flower with free ovule in place of stamen.

intermediate plants all had preponderately staminate racemes and were probably continuous variations of the normal type. Seed from one of these plants gave 202 individuals all of which were normal. A true intermediate plant, as will be shown later in the paper, always yields normal, intermediate and pistillate plants.

F₂ open-pollinated. (See Table 6).

TABLE 6

F₂ progeny of open-pollinated, normal plants; monoecious form 1.

Culture	%Germ	No. plants	No. N	No. I	No. P	%N	%I	%P
9N-1N	32.4	77	74	3	0	96.1	3.8	0
-2N	39.2	87	85	2	0	97.7	3.2	0
-3N	66.8	165	163	2	0	98.7	1.2	0
-4N	64.0	149	148	1	0	99.3	0.6	0
-5N	49.2	122	120	2	0	98.3	1.6	0
-6N	80.4	199	199	0	0	100.0	0.0	0

Culture	% Germ	No. plants	No. N	No. I	No. P	% N	% I	% P
-7N	38.4	93	92	1	0	98.9	1.0	0
-8N	62.4	149	145	4	0	97.3	2.6	0
-9N	69.2	166	166	0	0	100.0	0.0	0
-10N	40.4	101	101	0	0	100.0	0.0	0
-11N	59.6	148	148	0	0	100.0	0.0	0
-12N	69.6	172	172	0	0	100.0	0.0	0
-13N	23.6	58	58	0	0	100.0	0.0	0
-14N	42.8	103	100	3	0	97.0	2.9	0
-15N	62.4	152	152	0	0	100.0	0.0	0
-16N	58.4	146	146	0	0	100.0	0.0	0
-17N	52.4	119	117	2	0	98.3	1.6	0
-18N	66.2	152	152	0	0	100.0	0.0	0
-19N	75.2	179	178	1	0	99.4	0.5	0
-20N	83.2	192	190	2	0	98.9	1.0	0
-21N	65.6	155	154	1	0	99.3	0.6	0
-22N	76.8	176	175	1	0	99.4	0.5	0
-23N	38.4	91	91	0	0	100.0	0.0	0
-25N	36.8	79	79	0	0	100.0	0.0	0
Total	56.3	3230	3205	25	0	99.2	0.7	0

The F_2 generation presented in Table 6 was derived from 24 plants selected from culture 9N; each culture in the table represents the offspring from one seed parent. The selection was from culture 9N as it was the only one available at the time, the others recorded in Table 5 were grown in subsequent years. The results obtained in the F_2 generation were similar to the F_1 : 3,230 plants were grown; 99.2% normal; 0.7% intermediate, all bordering on the normal type and probably continuous variations; no pistillate plants were produced.

F_3 open-pollinated.

Four cultures derived from as many F_2 plants were grown: culture 9N-20N-1N gave 165 plants, all normal. Culture 9N-20N-1I was the progeny of an intermediate and consisted of 202 individuals, all normal. Culture 9N-23N-1N produced 194 plants, all normal. Culture 9N-25N-1N was derived from a normal plant which grew in a row contingent to pistillate strain 23P, all the seeds were planted and out of 1,204 individuals one was intermediate and all others normal.

Progeny of normal segregates from pistillate plants. (Monoecious form 2.)

Earlier in this paper data were given which showed that pistillate plants always produce some normal individuals; the percentage of these in open-pollinated cultures ranged from 1.7-58.0%, the average being 20.7%. These normal segregates are phenotypically identical to the true breeding normal strain. Because they do not breed true, as will be shown, they will be designated as monoecious form 2, and the true breeding strain, monoecious form 1, wherever it is necessary to distinguish between the two. It is significant that monoecious form 1 flowers earlier than the offspring of monoecious form 2. This must be taken into account in making crosses. For example, in cultures planted September 11, 1933, after eight weeks, 71% of the offspring of

form 1 were in flower, whereas, only 26% of form 2 were sufficiently mature for determination of floral types. The F_1 offspring of open-pollinated plants of form 2 are presented in Table 7.

TABLE 7
 F_1 progeny of open-pollinated, normal plants; monoecious form 2.

Culture	%Germ	No. plants	No. N	No. I	No. P	%N	%I	%P
23P-1N	84.8	193	85	76	32	44.0	39.3	16.5
-2N	76.0	173	85	59	29	49.1	34.1	16.7
-5N	74.4	171	95	51	25	55.5	29.8	14.6
-6N	78.4	185	95	68	22	51.3	36.7	11.8
-16N	56.4	129	61	44	24	47.2	34.1	18.5
81P-1N	46.4	109	65	36	8	59.6	33.0	7.3
-5N	80.0	194	55	110	29	28.3	56.7	14.9
-11N	71.5	139	58	79	2	41.7	56.8	1.4
150P-2N	11.6	25	13	11	1	52.0	44.0	4.0
-3N	11.2	23	16	6	1	69.5	26.0	4.3
-4N	—	118	91	22	5	77.1	18.6	4.2
23P-35P-1N	—	148	120	26	2	81.0	17.5	1.3
-2N	—	146	108	37	1	73.9	25.3	0.6
-3N	—	126	93	31	2	73.8	24.6	1.5
-4N	—	135	103	26	6	76.2	19.2	4.4
-5N	—	134	127	7	0	94.7	5.2	0.0
23P-38P-6N	—	99	57	28	14	57.5	28.2	14.1
-7N	—	96	47	34	15	48.9	35.4	15.6
-8N	—	95	57	32	6	60.0	33.6	6.3
-9N	—	93	62	23	8	66.6	24.7	8.6
-10N	—	96	56	32	8	58.3	33.3	8.3
Total		2627	1549	838	240	58.9	31.8	9.1

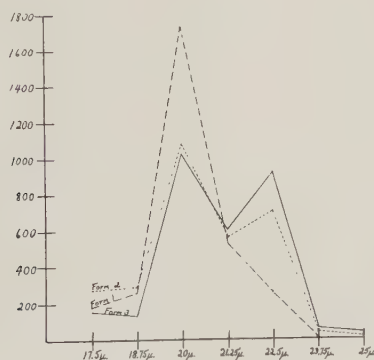


Fig. 11. Pollen measurements of monoecious form 1, 2 and 3.
Three thousand grains measured for each.

Monoecious form 2 is very clearly unlike monoecious form 1 in its genetical behavior. (Compare Table 5 and Table 7). The latter breeds true; the former usually yields pistillate, intermediate and normal plants. Cultures derived from form 2 had a preponderance of normal plants in every instance except 81P-5N and 81P-11N. However the frequency of each sex type in a

culture was extremely variable: normal plants ranged from 28.3-94.7%; intermediates from 5.2-56.8%; pistillate plants from 0.0-18.5%.

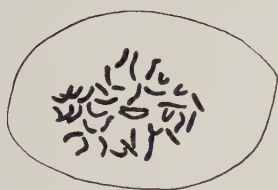
Culture 23P-35P-5N was unique in containing no pistillate plants. This suggested the possibility of deriving monoecious form 1 from this culture. Results have been favorable; one derivative has bred true for form 1 for two generations under open-pollination. Complete data will be given on this work because the result is evidence that the female parent entirely determines the sex type in a progeny.

A pistillate individual, 23P, produced 5% normal, 21% intermediate and 72% pistillate plants. One of the pistillate segregates, 23P-35P, yielded 58% normal, 39% intermediate and 2% pistillate plants. This is the highest percentage of normal individuals ever secured from a pistillate parent. Five of the normal segregates were selected and their progeny constitute cultures 23P-35P-1N to -5N in Table 7. These were predominantly normal cultures: -5N contained no pistillate plants. Four normal plants were selected from -5N and their offspring grown with the following results: Culture 1; 204 plants, all normal. Culture 2; 98 plants, all normal except for one intermediate which approached the pistillate type and was probably not a continuous variation of form 1. Culture 3; 203 plants, all normal. Culture 4; 206 plants, 199 normal, 5 intermediate and 2 pistillate. Progeny were then grown from the normal cultures 1 and 3. Offspring from culture 3 were not all normal: One culture from an open-pollinated plant consisted of 99 plants; 95 normal and 4 intermediate. Another culture, in which the normal plant was used as a female parent crossed to an intermediate, yielded 193 plants of which 180 were normal and 13 intermediate. Seed were sown from a plant of culture 1 and gave 100 plants all of which were normal. It will be interesting to see if successive generations of culture 1 remain true to type when open-pollinated or crossed to the intermediate type.

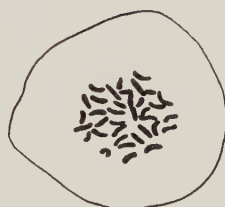
Monoecious form 1 and 2 in controlled pollinations. (See Table 8).

FIGURES

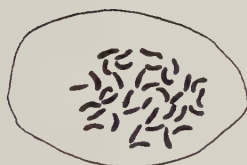
12. *A. elatior*, monoecious form 3; somatic prophase; 36 chromosomes.
13. *A. elatior*, pistillate type; somatic prophase; 36 chromosomes.
14. *A. elatior*, monoecious form 1; somatic prophase; 36 chromosomes.
15. *A. elatior*, monoecious form 1; metaphase I; 18 chromosomes.
16. *A. elatior*, monoecious form 1; metaphase II; 18 chromosomes.
17. *A. elatior*, monoecious form 2; somatic prophase; 36 chromosomes.
18. *A. elatior*, monoecious form 2; metaphase I; 18 chromosomes.
19. *A. elatior*, monoecious form 2; metaphase II; 18 chromosomes.
20. *A. trifida*; somatic prophase; 24 chromosomes.
21. *A. trifida*; metaphase I; 12 chromosomes.
22. *A. trifida*; metaphase II; 12 chromosomes.
23. *A. bidentata* x *A. trifida*; somatic prophase; 29 chromosomes.
24. *A. bidentata*; metaphase I; 17 chromosomes.
25. *A. bidentata*; metaphase II; 17 chromosomes.



12



13



14



15



16



17



18



19



20



21



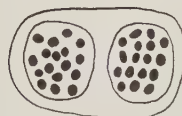
22



23



24



25

TABLE 8
Monoecious form 1 and 2 in controlled pollinations.

Culture	No. Plants	No. N	No. I	No. P	%N	%I	%P
1 selfed	106	105	0	1	99.0	0.0	0.9
2 selfed	536	287	203	46	53.5	37.8	8.5
F ₁ (1 x 2)	157	157	0	0	100.0	0.0	0.0
F ₂ (1 x 2)	30	30	0	0	100.0	0.0	0.0
F ₃ (1 x 2)	41	41	0	0	100.0	0.0	0.0
2 x 1	149	61	66	22	40.9	44.2	14.7
2 x 1 (repeated)	143	55	64	24	38.4	44.7	16.7
2 from (2 x 1) x 1	83	33	32	18	39.7	38.5	21.6
1 x 3	501	501	0	0	100.0	0.0	0.0
2 x 3	98	66	20	12	67.3	20.4	12.2
3 x 1	320	60	192	68	18.7	60.0	21.2
3 x 1 (repeated)	90	20	45	25	22.2	50.0	27.7
3 from (3 x 1) x 1	97	10	49	38	10.3	50.5	39.1
3 x 2	97	63	23	11	64.8	23.7	11.3
P x 1	235	42	95	98	17.8	40.4	41.7
P x 1 (repeated)	40	8	14	18	20.0	35.0	45.0

1: monoecious form 1.

2: monoecious form 2.

3: monoecious form 3.

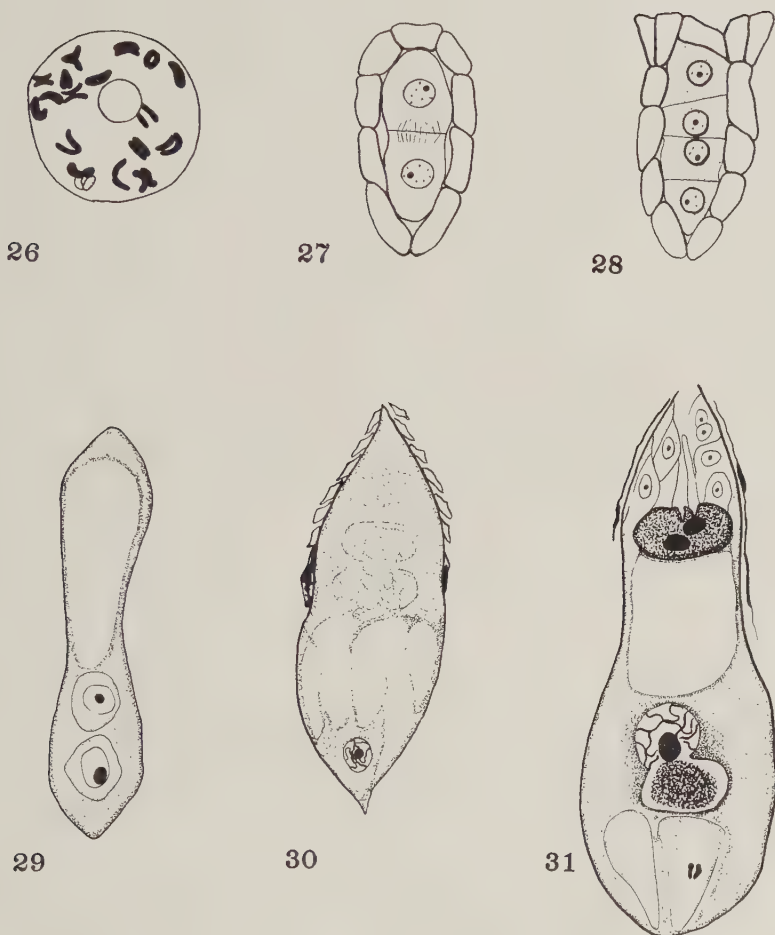
I: intermediate, monoecious form 3.

P: pistillate.

N: normal (1 or 2).

The data in Table 8 indicate that whenever monoecious form 1 is used as a female parent all offspring are normal regardless of the sex type of the male parent. This is in line with the more extensive work with open-pollinated plants in which form 1 bred true for three generations. Form 2, when used as a female parent, always gave a mixed progeny consisting of normal, intermediate and pistillate plants with the last type least abundant. This duplicated the behavior of form 2 under open-pollination.

Certain data in Table 8 require comment. The one pistillate plant secured from selfing form 1 was probably a contamination; it was the first plant in the culture and may have been misplaced; no other pistillate plant has arisen among several thousand individuals derived from form 1. The ratios from selfing form 2 happened to approach the average ratios under open pollination. In the latter case 2,627 plants consisted of 58.9% normal, 31.8% intermediate and 9.1% of the pistillate type. It may seem peculiar that form 2, when used as a female parent, gave the highest percentage of normal individuals when crossed with the intermediate type. This does not mean that intermediate plants are prevailingly normal in their genetical behavior; it will be shown that they behave quite otherwise when used as the female parent and that all genotypes behave alike when used as the male parent. The writer cannot definitely explain the high percentage of normal plants in this instance. However, individuals of form 2 may give quite different ratios when used as seed



FIGURES

26. *A. elatior*, pistillate type; diakinesis in megaspore mother cell; 18 pairs of chromosomes.
27. *A. elatior*, pistillate type; first division of megaspore mother cell.
28. *A. elatior*, pistillate type; tetrad of megaspores.
29. *A. elatior*, pistillate type; two-celled embryo sac.
30. *A. elatior*, pistillate type; four-celled embryo sac; nucellus degenerating.
31. *A. elatior*, pistillate type; mature embryo sac; several antipodals, triple fusion nucleus irregularly shaped, fusion of sperm and egg, two synergids; nucellus fragmentary.

parents and the environment can alter these ratios as will be demonstrated in another paper. The cross in question, 2×3 and its reciprocal, gave similar, aberrant ratios; both were made at the same time with closely related plants during a different year than the other crosses were made.

C. THE GENETICAL BEHAVIOR OF THE INTERMEDIATE TYPE; MONOECIOUS FORM 3.

F_1 open-pollinated.

The progeny of wild, open-pollinated, intermediate plants were grown with the following results. Cultures 41, derived from one plant; 414 plants, 224 or 54.1% normal, 164 or 39.6% intermediate, and 26 or 6.2% pistillate. In addition ten cultures (3,720 plants) were grown from which incomplete data were secured. All of these cultures consisted of three sex types; normal plants comprised 17-56% of a culture.

In a culture from seed collected at Fayetteville, Arkansas, by Professor Delbert Swartz, three intermediate plants occurred among 229 plants of the normal type. One of the intermediate plants bordered on the pistillate type. All of its seed were sown with the following results: 455 plants; 125 or 27.4% normal; 330 or 72.5% intermediate; no pistillate plants. This intermediate progenitor behaved unlike any of local origin; it gave no pistillate offspring and had the highest percentage of intermediate plants ever secured. Most of the intermediates approached the normal type; only 28 were predominantly pistillate. The culture was conspicuously vigorous and flowered a month later than culture O2N (Table 5) which was derived from a normal plant in the same population.

The progeny of intermediate segregates from the pistillate type.

Pistillate plants always produce some intermediate individuals; the percentage of these in cultures under open-pollination has ranged from 23.1-58.7%, the average 39.0%. The results of growing progeny of intermediate segregates from pistillate plants are recorded in Table 9.

TABLE 9

F_1 progeny of intermediate segregates from the pistillate type; monoecious form 3.

Culture	% Germ	No. plants	No. N	No. I	No. P	% N	% I	% P
23P-1I	41.2	97	47	32	18	48.4	32.9	18.5
-2I	46.0	111	25	58	28	22.5	52.2	25.2
-10I	42.8	100	48	31	21	48.0	31.0	21.0
150P-1I	36.8	84	19	39	26	22.6	46.4	30.9
-2I	57.2	133	61	50	22	45.8	37.5	16.5
-3I	65.6	145	40	50	55	27.5	34.4	37.9
23P-38P-11I	—	91	35	41	15	38.4	45.0	16.4
-12I	—	94	27	47	20	28.7	50.0	21.2
-13I	—	87	30	43	14	34.4	49.4	16.0
-14I	—	96	38	45	13	39.5	46.8	13.5
-15I	—	95	31	48	16	32.6	50.5	16.8
Total		1133	401	484	248	35.3	42.7	21.8

Each culture in Table 9 was derived from a single intermediate plant; three sex types occurred in each culture but in widely varying frequencies. In seven of the cultures the intermediate type predominated; in three cultures the normal type was most abundant, and in only culture 150P-3I the pistillate type occurred in highest frequency.

It was pointed out in the introduction that intermediate plants are extremely variable phenotypically. They range from plants which would be normal but for one pistil in one androgynous head to plants which would be pistillate but for one staminate flower. Since individual intermediate plants give variable ratios in their progeny it might be expected that predominantly pistillate plants would give a higher percentage of pistillate offspring than would predominantly normal plants. The limited data do not support this assumption. Seed were collected from five open-pollinated, intermediate plants in the same culture, 23P-38P. Three of these were predominantly pistillate and their offspring (Table 9: cultures 23P-38P-13I, 14I-15I) together yielded 278 plants; 35.6% normal, 48.9% intermediate and 15.4% pistillate. Two of the intermediate plants were predominantly normal and their offspring (Table 9; cultures 23P-38P-11I and -12I) together yielded 185 plants; 33.5% normal, 47.0% intermediate and 18.9% pistillate. These ratios are certainly similar; the predominantly pistillate plants, oddly, gave slightly fewer pistillate individuals than did the predominantly normal plants.

Table 10 gives a résumé of the behavior of the three monoecious forms and the pistillate type under open-pollination.

TABLE 10
Résumé of genetical behavior of three monoecious forms and pistillate type under open pollination.

Female parent	No. plants	No. & % N	No. & % I	No. & % P	Range N	Range I	Range P
1	7,521	7,491 or 99.6%	30 or 0.39%	0.0 or 0%	96.1 to 100%	0 to 3.8%	0 to 0%
2	2,627	1,549 or 58.9%	838 or 31.8%	240 or 9.1%	28.3 to 94.77%	5.2 to 56.8%	0 to 18.5%
3	1,547	625 or 40.4%	648 or 41.8%	274 or 17.7%	22.5 to 54.1%	31.0 to 52.2%	6.2 to 37.9%
P	6,798	1,426 or 20.9%	2,652 or 39.0%	2,720 or 40.0%	1.7 to 58.0%	23.1 to 58.7%	2.0 to 72.8%

A massing of all data on open-pollination (Table 10) emphasizes three facts: (1) Monoecious form 1 breeds true irrespective of the pollen parent. (2) On the average the percentage of pistillate plants in a progeny is highest if the female parent is of pistillate type and decreases in the following order; female parent monoecious form 3, form 2, and form 1. (3) Individuals of a

given sex type when used as the female parent give extremely variable ratios of the sex types in their progeny (form 1 excepted).

The intermediate type (monoecious form 3) in controlled pollinations (Table 11).

TABLE 11
The intermediate type, form 3 monoecious, in controlled pollinations.

Culture	No. Plants	No. N	No. I	No. P	%N	%I	%P
3 selfed	313	72	153	88	23.0	48.8	28.1
3 x 1	320	60	192	68	18.7	60.0	21.2
3 x 1 (repeated)	90	20	45	25	22.2	50.0	27.7
3 from (3 x 1) x 1	97	10	49	38	10.3	50.5	39.1
1 x 3	501	501	0	0	100.0	0.0	0.0
3 x 2	97	63	23	11	64.8	23.7	11.3
2 x 3	98	66	20	12	67.3	20.4	12.2
P x 3	274	44	114	116	16.0	41.0	42.3
P x 3 (repeated)	96	12	42	42	12.5	43.7	43.7

The results in Table 11 are in keeping with the more extensive work in the field under open-pollination. The intermediate type when used as a female parent, regardless of the sex type of the male parent, throws all sex types, with the intermediate individuals usually predominating. When the intermediate type is employed as a male parent the results differ from the foregoing, depending entirely on the sex type of the female parent. Throughout the investigation results indicated that the proportion of sex types in a progeny was controlled entirely by the female parent; pollen from all plants seemed to behave alike and to contribute no factors for the expression of monoecious form 2, 3 or pistillate plants. A study of the pollen grains and of the chromosomes was accordingly undertaken.

d. POLLEN STUDIES

Pollen grains were examined to detect possible peculiarities which might indicate why pollen of all sex types functions alike, i.e. seems never to carry factors for the expression of monoecious form 2, 3 or the pistillate type. There was no significant percentage of aborted pollen; each sex type produced less than 10% of aborted grains. The only striking peculiarity was the diversity in the size of the pollen grains; this was true of pollen from all plants so that it was necessary to make extensive measurements in order to evaluate its possible significance. A random sample of 200 pollen grains was measured from each of 15 plants of each sex type; monoecious form 1, 2, and 3. (A total of 3,000 measurements for each sex type).

The results are given graphically in Fig. 11. The measurements, in microns, are charted along the abscissa and the frequencies along the ordinate. Monoecious form 1, which breeds true for the normal type, when used as a female parent, gave essentially a normal frequency curve; the mode was 20 microns and the mean 20.14 microns. Monoecious form 2, which gives three sex types when used as a female parent, with an average of about 59% normal plants, gave a bimodal curve; the main mode 20 microns, another at 22.5

microns and the mean 20.54 microns. Form 2 had 772 grains measuring 22.5 microns or more, whereas, form 1 had only 268. Pollen measurements for monoecious form 3 gave even a more marked bimodal curve; the modes were again at 20 microns and 22.5 microns; the mean was 20.92 microns. There were 1,025 pollen grains which measured 22.5 microns or more.

The characteristic of producing large pollen grains is definitely possessed by the sex types which, when used as female parents, yield pistillate segregates. Form 1 does not produce pistillate plants and produces relatively few large pollen grains. Form 2 and 3 yield pistillate segregates; both produce a high number of large pollen grains and form 3 which yields more pistillate plants has the higher frequency of large grains. It is reasonable to assume that the large pollen grains carry factors for the expression of the pistillate type and that for some reason these grains do not function as this type is never transmitted through the male parent. The writer has no direct evidence to prove this assumption. Pollen grains of *Ambrosia elatior* L. have not been successfully germinated on some 40 nutrient media, yeast extract, stigma extracts and excised stigmas. Some technique must be devised to clear this important point.

e. CYTOLOGICAL STUDIES

A variation in the size of pollen grains has often been found to be proportional to chromosome number. In tulips, for example, de Mol (1928) found that the monoploid, diploid and tetraploid pollen grains show a volume ratio of 8:27:64.

Although the unusual genetical behavior and the striking differences in pollen grains in *Ambrosia elatior* L. strongly suggest diverse chromosome sets for separate sex types, my studies, up to the present, have not demonstrated chromosome differences. The $2n$ number is 36 for all sex types and the n number 18 (Figs. 12 to 19). Meiosis proceeds without any unusual vicissitudes. The chromosomes are unfortunately very small and it is perfectly possible that there are morphological peculiarities which could be detected by an experimenter endowed with a nicer technique and capable of more critical observations.

Other species of *Ambrosia* have been cultured and, as a matter of record, have been found to possess the following chromosome numbers: *A. trifida* (Figs. 20, 21, 22) $2n=24$ chromosomes, $n=12$ chromosomes. *A. bidentata* (Figs. 24, 25) $n=17$. *A. bidentata* $\times$ *trifida* (Jones, 1933) (Fig. 23) $2n=29$.

Embryo sacs were studied to check on apomixis which is rather common in the *Compositae*. All sex types have been kept emasculated during the winter season, and, although they bore numerous pistillate heads, produced no seed. It is possible that pollination stimulates an apomictic development, perhaps a nucellar budding or parthenogenetic development of a diploid egg. It must be admitted that the vegetative diversity within each sex type is so great that such a possibility does not seem likely.

The development of the embryo sac has been studied in all sex types; it

is essentially normal as are the subsequent fertilization, embryo and endosperm formation. It is not easy to study the chromosomes during megasporogenesis; there is only one megaspore mother cell in a pistil and chromosome counts have been possible only at diakinesis. Fig. 26 illustrates a megaspore mother cell of a pistillate plant in the diakinesis stage. In eight instances the chromosomes are paired laterally; four bivalents are formed by chromosomes joined end to end; the members of one pair merging with the nucleolus are not in contact and could be considered as two univalents; there are five instances where only a single structure is seen, but these are, in the opinion of the writer, too large to be univalents and are believed to be bivalents in which the members are closely paired laterally.

Interkinesis following the first meiotic division is illustrated in Fig. 27 and then in turn: tetrad of megaspores (Fig. 28), two-celled embryo sac (Fig. 29), four-celled embryo sac (Fig. 30) and the mature embryo sac with synergamy occurring (Fig. 31). The nucellus degenerates early and the mature embryo sac is neatly nestled within a specialized layer of the heavy integument. As in many Compositae, (Afzelius, 1924; Schnarf, 1931) the antipodal apparatus consists of several cells. Fusion of the polar nuclei has been observed. Fig. 31 shows what is probably the endosperm nucleus shortly after triple fusion; the outline is very irregular and two large nucleoli are present.

Discussion

Ambrosia elatior L. is one of many species producing pistillate plants and also plants bearing both stamens and pistils. Often the floral types in these species have been found to be inherited but the breeding results have been complex and difficult to formulate. Undoubtedly the manner of inheritance would be clearer if the general genetical behavior for the species were better understood. The genes for floral types, and there may be many genes of graded potencies in several chromosomes, cannot very well be allocated and their phenotypic expressions traced if few or no genes for vegetative characteristics have been charted.

Zea Mays is unique in having many genes mapped. As a result the inheritance of floral types, though sometimes complex, can be definitely assigned to the activity of certain genes, e.g.:

Four recessive genes are each capable of producing pistillate plants; three or more recessive genes can each give rise to strains in which the individuals produce only a few or no pistillate flowers; two recessive genes are potentially able to influence the formation of two types of andromonoecious strains. (Emerson, 1924.)

If anyone feels skeptical he needs but realize that using such information it has been possible to create dioecious maize. Jones (1934) has developed one type by bringing into combination two specific sex-influencing genes whose loci are known. One of these is kept in the heterozygous condition in the male, the other is homozygous in both sexes. The interaction of both is necessary to maintain the unisexual condition. Emerson (1932) has developed two types of dioecious maize; in one of these the male is heterozygous and in the other the female.

There are monoecious species in which the sex condition is the expression of a simpler gene complex. Pastrana (1932) has made the remarkable discovery in *Begonia Schmidtiana* of the presence of an unpaired chromosome which fails to enter the stem initial of a staminate flower which consequently has one less chromosome than the pistillate flower; the unpaired chromosome is probably concerned with the determination of sex. Rosa (1927) has found that in *Cucumis melo*, *C. sativus*, and *Citrullus vulgaris*, in which there are monoecious and andromonoecious varieties, the monoecious type depends upon a single dominant gene.

Sometimes the inheritance of sex does not permit a gene analysis because of the nature of the material. Hermaphrodites occur among dioecious species in which they behave as though derived from one sex. Shull (1910) found certain hermaphrodites in *Lychnis dioica* to behave like modified staminate plants. It seems to the writer that Newton's (1931) results on *Silene Otites* could be similarly explained. Here pistillate x hermaphrodite gave pistillate and staminate plants in about equal proportions, together with a very small percentage of hermaphrodites; hermaphrodite x staminate produced cultures predominantly staminate. Rosa (1925) considers the monoecious forms of spinach to be modified pistillate plants.

Numerous examples have been reported in which progeny tend to have the floral type of the female parent; this is often more pronounced the closer the female parent approaches the pistillate type. (*Mercurialis annua*, Yampolsky, 1930; *Satureja hortensis*, *Silene inflata* and *Plantago lanceolata*, Correns, 1905, 1906, 1908). Correns (1908) found that in *Plantago lanceolata* strong females crossed with hermaphrodites gave a prevailing pistillate progeny. However, by using pollen from different hermaphrodites on the same pistillate plant, it was demonstrated that the pollen parent had some influence on the proportion of sex forms in the progeny. Bartlett (1913) found that the pollen parent had equal influence in crosses between certain hermaphrodites of *P. lanceolata*.

Ambrosia elatior, like many of the foregoing species, has not been subjected to a general genetical analysis. It must be admitted that the inheritance of floral types, as presented in this paper, is sufficiently irregular to warrant an interpretation based on protoplasmic lability resulting in graded potencies of sex cells and consequently various graded sex expressions, such as in *Mercurialis annua* (Yampolsky, 1930). However, it seems to the writer, that the inheritance is particulate enough to hazard a tentative genetical interpretation.

Briefly, this is the proposed genetical interpretation. The expression of each of the four sex forms is controlled by the action of many genes, of differing potencies, located in several chromosomes. Monoecious form 1 is so strongly differential for its type that it behaves as though sexually homozygous. Functional male gametes of all sex forms possess the same gene complex; differential for the expression of form 1. The other forms are sexually very heterozygous. Usually a pistillate plant has a gene complex more differential for the expression of the pistillate form than has a plant of monoecious form 3 which in turn exceeds form 2. The gene complex of a male gamete, differ-

ential for form 1, may or may not be overbalanced by the pistillate complex of a female gamete because the latter will vary widely. The three forms, 2, 3 and pistillate are not to be regarded as the expression of only three definite genotypes; many genes are involved and consequently many imperceptibly graded genotypes, some strongly differential for a particular phenotypic expression, others capable of developing into any of the three sex forms depending upon the interaction with the environment.

The following evidence is given in support of the tentative genetical scheme:

(a) All functional pollen has the same effect on the proportion of sex forms in a progeny. (Therefore, all male gametes have the same gene complex for the expression of floral type.)

1. Monoecious form 1 breeds true regardless of the sex form of the male parent. This sex form was grown for three generations under open-pollination; 7,521 plants were grown and 99.6% were of form 1. Controlled crosses gave the same results. There is no evidence of apomixis.

2. A given sex form employed as a female parent gives similar ratios with different pollen parents. The data are presented in Table 12.

TABLE 12

Female parent	Male parent	No. plants	% pistillate
1	1	105	0.9
1	2	157	0.0
1	3	501	0.0
2	2	536	8.5
2	1	149	14.7
2	3	98	12.2
3	3	313	28.1
3	1	90	27.7
3	2	97	11.3
P	1	235	41.7
P	3	274	42.3

1, 2, and 3 refer to monoecious forms 1, 2 and 3; P to pistillate.

The data support the point in question except for (3 x 2) which gave far too few pistillate plants. These results were not obtained by crossing a single female parent with two pollen parents. This would be more confirmatory as plants within a sex form give variable ratios when used as a female parent. The writer has since worked out a method whereby one female parent can be so tested.

3. A strain, 23P-35P-5N, breeding true for form 1, has been developed from form 2 by selection under open-pollination. It seems unlikely that the homozygous form 1 could be so derived in a wind pollinated species if functional pollen grains carried different gene complexes for sex expression.

b. Functional pollen grains contribute only to the expression of monoecious form 1.

1. Monoecious form 1 breeds true to type regardless of the sex form of the pollen parent. If any pistillate factors are contributed by the pollen other sex forms should segregate from form 1.

2. If male gametes contribute no pistillate genes all gene complexes should become less pistillate in succeeding generations. For example, pistillate plants vary in the number and potency of their pistillate genes although they are phenotypically alike; in succeeding generations they should have fewer pistillate genes and therefore yield, in turn, fewer pistillate offspring. Data under open-pollination confirm this: pistillate plant 23P produced 531 plants of which 72.8% were pistillate. Twenty-one pistillate plants selected from the F_1 yielded 3,046 plants of which 41.1% were pistillate and no progeny from a single plant gave as great a proportion of pistillate individuals as the F_1 . Five pistillate plants selected from the F_2 yielded 488 plants of which 31.6% were pistillate. Successive generations of monoecious form 2 agree with these results. There are no data for form 3. The relative scarcity of pistillate plants in nature is also significant; certainly these have greater survival value than all other forms which they greatly exceed in vegetative vigor and in seed production.

(c) The percentage of pistillate plants produced by the sex forms is usually in the following order: pistillate, monoecious form 3, form 2 and form 1. (Therefore the gene complexes have the same order for pistillate expression.)

The average percentage of pistillate plants under open pollination has been as follows: Pistillate form 6,798 plants, 40.0% pistillate; monoecious form 3 gave 1,547 plants, 17.7% pistillate; form 2 gave 2,627 plants, 9.1% pistillate; form 1 gave 7,521 plants, 0.0% pistillate. Results under controlled pollination were similar as Table 12 shows.

Most cultures conform roughly to the averages just given. However, individual cultures, each derived from a single plant, can be selected which yield very different results and this is a very important consideration. For example: pistillate plant 23P-35P gave 198 plants, 2.0% pistillate; monoecious form 3, plant 4I, gave 414 plants, 6.2% pistillate; form 2, plant 23P-16N, gave 129 plants, 18.5% pistillate. Here the order of pistillateness is the reverse of the foregoing and yet the cultures were sizeable.

The variability in breeding behavior and experimental evidence that the environment may influence the ratios indicate that monoecious form 2, 3 and pistillate are the expressions of numerous, imperceptibly graded genotypes. Some of the genotypes must be relatively fixed in their phenotypic expression or the average breeding results for the three sex forms in question would not be so dissimilar. Other of the genotypes must be capable of developing into any of the three sex forms depending on the interaction with the environment.

(d) Individual plants of one sex form show dissimilar ratios when pollinated by homozygous form 1. (Therefore, plants of one sex form vary in their genotypes; the gene complexes are multiple and individual genes influencing sex vary in potency.)

Five pistillate plants were pollinated by form 1 with the following results: plant no. 20 produced 19 offspring, 10.5% pistillate; no. 31 produced 57 offspring, 26.3% pistillate; no. 26 produced 18 offspring, 38.8% pistillate; no. 24 produced 37 offspring, 54.0% pistillate; no. 17 produced 104 offspring, 51.9% pistillate.

Summary

1. Characteristics which have been used to establish *Ambrosia elatior* L. and *A. artemisiifolia* L. as separate species are too variable in culture to justify segregation. *Ambrosia elatior* L. should be retained as it has priority of position in the Species Plantarum.

2. *Ambrosia elatior* L., in nature, is usually monoecious; there are occasional pistillate plants and plants which intergrade between the monoecious and pistillate habit. The monoecious plants, for convenience in breeding experiments, are divided in three categories.

3. Monoecious form 1, which is the prevalent form in nature, breeds true when used as a female parent. The sex form of the pollen parent in *A. elatior* never affects the nature of the offspring.

4. Monoecious form 2, which is phenotypically identical to form 1, when used as a female parent, gives on the average 10% pistillate plants.

5. Monoecious form 3, which is intermediate between form 2 and pistillate, when used as a female parent, gives on the average 20% pistillate plants.

6. Pistillate plants give on the average 40% pistillate offspring.

7. Individual cultures, with the exception of form 1, may vary widely from these averages.

8. All sex types are interfertile and all monoecious forms self fertile. There is no evidence of apomixis.

9. There is a correlation between the production of large pollen grains, presumably non-functional, and the tendency of a sex form to produce pistillate offspring.

10. The n chromosome number of all sex types is 18 and the $2n$ number 36. There are no constant, visible differences in the chromosomes correlated with a given sex type.

11. The breeding behavior could probably be explained by a theory of protoplasmic lability resulting in graded potencies of sex cells. However, the writer favors a multiple gene interpretation because the inheritance seems to him to be particulate.

The author wishes to thank Professor H. H. Bartlett for help he has given in this study.

REFERENCES

- AFZELIUS, K. 1924—Embryologische und zytologische Studien in *Senecio* und verwandten Gattungen. *Acta horti Bergiani* 8:123-219.
- BARTLETT, H. H. 1913—Inheritance of sex forms in *Plantago lanceolata*. *Rhodora* 15:173-178.
- CORRENS, C. 1905—Weitere Untersuchungen über die Gynodioecie. *Ber. Deut. Bot. Gesell.* 23:452-463.
- 1906—Die Vererbung der Geschlechtsformen bei den gynodiöcischen Pflanzen. *Ber. Deut. Bot. Gesell.* 24:459-472.
- 1908—Die Rolle der männlichen Keimzellen bei der Geschlechtsbestimmung der gynodiöcischen Pflanzen. *Ber. Deut. Bot. Gesell.* 26a:686-701.
- DAVIS, W. E. 1930—Primary dormancy, after-ripening, and development of secondary dormancy in embryos of *Ambrosia trifida*. *Amer. Jour. Bot.* 17:58-77.
- EMERSON, R. A. 1924—A genetic view of sex expression in the flowering plants. *Science* 59:176-182.
- 1932—The present status of maize genetics. *Proc. Sixth Int. Congress Genetics* 1:141-152.
- GARNER, W. W., AND H. A. ALLARD. 1920—Effect of the relative length of day and night and other factors of environment on growth and reproduction in plants. *Jour. Agr. Res.* 18:553-606.
- JONES, D. F. 1934—Unisexual maize plants and their bearing on sex differentiation in other plants and in animals. *Genetics* 19:552-567.
- JONES, K. L. 1933—*Ambrosia bidentata* Michx. x *A. trifida* L. *Amer. Midl. Nat.* 14:720-724.
- LINNAEUS, C. 1753—*Species Plantarum*.
- MOL, W. E. DE. 1928—Producing at will of fertile diploid and tetraploid gametes in Duc von Thol, Scarlet (*Tulipa suaveolens* Roth). *Vierteljahrsschr. Naturf. Gesell. Zürich.* 73(Beibl. 15):73-97.
- NEWTON, W. C. F. 1931—Genetical experiments with *Silene Otites* and related species. *Jour. Genetics* 24:109-120.
- PASTRANA, M. D. 1932—Sporogenesis and sex determination in *Begonia Schmidtiana*. *Amer. Jour. Bot.* 19:365-384.
- ROSA, J. T. 1925—Sex expression in spinach. *Hilgardia* 1:259-274.
- 1927—The inheritance of flower types in *Cucumis* and *Citrullus*. *Hilgardia* 3:233-251.
- RYDBERG, P. A. 1922—*Carduales*. *North Amer. Flora* 33:1-46.
- SCHNARF, K. 1931—*Vergleichende Embryologie der Angiospermen*. 354 pp. Berlin.
- SHARP, L. W. 1924—The factorial interpretation of sex-determination. *La Cellule* 35:195-235.
- SHULL, G. H. 1910—Inheritance of sex in *Lychnis*. *Bot. Gaz.* 49:110-125.
- STEVENS, O. A. 1932—The number and weight of seeds produced by weeds. *Amer. Jour. Bot.* 19:784-794.
- YAMPOLSKY, C. 1930—The further behavior of sex in *Mercurialis annua*. *Ztschr. indukt. Abstam. u. Vererbungslehre.* 55:267-299.
- 1934—The cytology of the intersexual flowers of *Mercurialis annua*, a morphogenetic study. *Amer. Jour. Bot.* 21:651-673.

